Controlling E. coli O157

G.D. Betts

Introduction

Escherichia coli is a bacterium that exists naturally as part of the normal gut flora of healthy humans and other mammals. The majority of strains are harmless and do not cause any food poisoning problems; however, there are a few strains of this organism which are pathogenic to humans and are associated with food related illness.

One such strain of E. coli O157 is a particularly virulent food poisoning organism that only requires a small number of cells to cause illness. The exact infective dose is unknown but there have been a number of food poisoning cases where the infected foods have been found to contain low contamination levels and some estimates state that as few as ten cells are required to cause illness. E. coli O157 was first isolated from foods in Canada in 1974 and was recognized as a foodborne pathogen in 1982 (Riley et al., 1982) following an outbreak of gastroenteritis in the USA involving 42 people who had eaten hamburgers. Food poisoning from E. coli O157 results in a number of symptoms ranging from mild diarrhoea to the more serious haemolytic uraemic syndrome which can result in kidney failure and ultimately death. Over the past decade, the numbers of reported food poisoning cases due to E. coli O157 have risen steadily in England and Wales from 385 cases in 1993 to 1,102 cases in 1999 (Anonymous, 2000).

Foods associated with E. coli disease have primarily been meat and meat products including beef and lamb, but other products have been implicated, including cheese, lettuce and fruit products. A particular problem for food manufacturers is that this organism may survive in acidic products, for example, fermented meats, apple juice and mayonnaise. Studies were done to evaluate the growth and survival of E. coli O157 in acid environments. It was shown that this organism could grow at a pH of 3.75 and could survive for seven days at pH 3.3 and six hours at pH 2.8 in the presence of 3 per cent (v/v) acetic acid. These data are of use to manufacturers of acid products to control the growth and survival of E. coli O157.

Preventing growth of E. coli

E. coli O157 is not a superbug and has very similar growth and survival characteristics to

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Keywords

E. coli, Principal Research Officer, CCFRA, Food, Food poisoning

Abstract

Escherichia coli O157 is a virulent food pathogen that can cause severe illness in humans. The number of cases is steadily rising and reached over 1,000 in 1999. Foods associated with E. coli O157 are primarily meat and meat products, but other foods have been implicated, including cheese, lettuce and fruit products. A particular problem for food manufacturers is that this organism may survive in acidic products, for example, fermented meats, apple juice and mayonnaise. Studies were done to evaluate the growth and survival of E. coli O157 in acid environments. It was shown that this organism could grow at a pH of 3.75 and could survive for seven days at pH 3.3 and six hours at pH 2.8 in the presence of 3 per cent (v/v) acetic acid. These data are of use to manufacturers of acid products to control the growth and survival of E. coli O157.

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other enteric organisms such as non-O157 E. coli and Salmonella. It survives freezing at −20°C and can survive chill storage, being able to grow down to a minimum temperature of 6.5°C (Anonymous, 1997) which is similar to that for non-pathogenic E. coli (7°C) or Salmonella (7°C). E. coli O157 can also tolerate similar salt levels to other typical pathogens, e.g. it can grow at a water activity as low as 0.95 (equivalent to 8 per cent salt).

Thermal inactivation studies have shown that E. coli O157 is no more heat resistant than typical Salmonella and Listeria species and therefore food pasteurisation processes for chilled foods (e.g. 70°C for two minutes), which are designed to eliminate Listeria spp. should also be sufficient with respect to this organism.

One particularly notable characteristic of E. coli O157 is the reported high resistance to acid (Bennett and Betts, 1997). It has been shown that E. coli O157 can survive in a range of acidic environments, although the exact growth and survival characteristics will depend on acid type and strength used.

In order to gain a better understanding of the acid conditions required to inhibit the growth of this organism, some work was done at CCFRA to investigate the minimum pH and temperature conditions which allowed or prevented growth of E. coli O157 using a range of acid types (Gonzalez et al., 1999). Table I shows the combinations tested and the conditions which allowed growth within 30 days.

Table I Minimum pH allowing growth of E. coli O157 within 30 days in different acid conditions

<table>
<thead>
<tr>
<th>Acid Type</th>
<th>pH</th>
<th>8°C</th>
<th>12°C</th>
<th>15°C</th>
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<tbody>
<tr>
<td>Lactic acid</td>
<td></td>
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<tr>
<td>pH 3.56</td>
<td>±</td>
<td>±</td>
<td>±</td>
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<td>pH 3.78</td>
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<td>+</td>
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<td>pH 4.02</td>
<td>±</td>
<td>+</td>
<td>+</td>
<td>+</td>
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<td>pH 4.24</td>
<td>±</td>
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<td>pH 4.45</td>
<td>±</td>
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<td>+</td>
<td>+</td>
</tr>
<tr>
<td>Citric acid</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>pH 3.75</td>
<td>±</td>
<td>±</td>
<td>+</td>
<td>+</td>
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<tr>
<td>Acetic acid</td>
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<td>pH 3.64</td>
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<td>pH 4.32</td>
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<tr>
<td>pH 4.52</td>
<td>±</td>
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</tbody>
</table>

Notes: − = no growth; + = growth

It can be seen that the storage temperature had a marked effect on the growth of E. coli O157 in acidic environments. No growth was observed at the lowest temperature of 8°C even under the relatively mild acid environment of pH 4.5. As 8°C is close to the minimum reported growth temperature for this organism, it would appear that this organism is not able to tolerate the low pH conditions in combination with low temperature. This gives some useful information for food producers where a combination of antimicrobial treatments may be needed to inhibit growth of this organism.

When the temperature was increased to 15°C, growth was observed at a pH of 3.75 when citric acid was used to reduce the pH. At these temperatures, low pH alone may not be sufficient to prevent growth of E. coli O157 if the acid type used is weakly antimicrobial.

The data in Table I also show the importance of the acidulant used in product formulation.

Acetic acid had by far the greatest effect on growth of E. coli O157 and no growth was observed under any condition tested. Products such as mayonnaise and pickles that use acetic acid should therefore be able to prevent the growth of E. coli O157, whereas products containing more citric acid, e.g. fruit juice products, may not have such a protective environment.

Controlling the growth of E. coli O157 is just one aspect of importance with respect to this organism. As this organism has a very low infective dose, an important factor to consider with respect to food safety is not simply whether this organism can grow in a product but whether it can survive during storage, as contamination with, and survival of, a few cells may be sufficient to cause food poisoning.

### Survival of E. coli in foods

Many of the normal human gut flora are able to survive for extended periods of time in acidic environments and E. coli O157 also possesses this characteristic.

Recently the survival of E. coli O157: H7 has also been studied in orange juice stored at refrigeration temperatures (3°C) (Linton et al., 1999), and it was shown that at pH 4.5 there was very little decrease in levels of this organism during 26 days’ storage. Even when
the pH was reduced to 3.4, detectable levels of *E. coli* were present up to 20 days. Survival has also been reported in salami, mayonnaise and fruit juices and outbreaks of food poisoning have been linked to these products.

In order to assess the survival of *E. coli* O157 in acidic environments a study was done (Gonzalez *et al.*, 1999) to evaluate the effect of different storage temperatures (8, 12 and 15°C) and level of organic acid (0, 1 and 3 per cent) at pH 2.8 and 3.3. The aim of this study was to evaluate the likely survival time of this organism in acid foods and drinks. Different laboratory media were inoculated with approximately $10^6$ *E. coli* O157 cells per ml and evaluated for changes in numbers during storage.

The results from this study showed that the type and level of organic acid had a significant effect on the survival of *E. coli* O157 whilst the temperature had little effect. The greatest survival time was at 0 or 0.5 per cent acetic acid and a pH of 3.3, when it took over seven days for the levels of *E. coli* O157 to become undetectable. When the level of organic acid used was increased to 3 per cent (v/v), the survival time of *E. coli* was reduced from seven days to six hours. This is shown in Figure 1 for *E. coli* O157 at pH 3.3, 22°C and 0 to 3 per cent (v/v) acetic acid.

These data show that *E. coli* can survive for extended periods of time in products with a pH of 2.8-3.3 and an acetic acid concentration of 1 per cent (v/v). These data have relevance for foods that are naturally high in organic acids or are formulated to contain these acids.

The proportion of different organic acids present in such food will depend on the manufacturer’s formulation and for fruit and vegetables will depend on the variety used or even the particular season. In general, the level of organic acids in foods can total 3 per cent in fruits and between 0.1 and 0.8 per cent in vegetables (Holland *et al.*, 1991). Fresh orange juice contains 1 per cent citric acid and in commercial mayonnaise and dressings the acetic acid level can range from 0.3 to 0.7 per cent and from 0.03 to 1.34 per cent (Muriana and Kanach, 1995). All such products fall within the range tested above and the data could therefore be used to help control the survival of *E. coli* O157 during formulation of these products.

**Effective disinfection of *E. coli* O157**

It has been shown that *E. coli* O157 has the potential to survive in acidified products for several weeks. Owing to the small infective dose of this organism, even a small number of surviving cells could cause food safety problems. It is therefore important to minimise the potential for the presence of this organism in foods. One way to do this is to ensure that effective disinfectants or sanitisers are used.

Taylor *et al.* (1999) evaluated the effectiveness of 18 disinfectants commonly used in the food industry against two organisms: *Pseudomonas aeruginosa*, a common disinfection test organism, and *E. coli* O157: H7. Tests were done at 10 and 20°C according to the requirements of the BS EN 1276 (1997) quantitative resuspension test for the evaluation of the bactericidal activity of chemical disinfectants for the food industry.

Bacterial suspensions were added to various concentrations of the disinfectants and left for a contact time of five minutes, after which the number of surviving bacteria were calculated. For a disinfectant to pass this test it must achieve a 5-log reduction in numbers of the test organism. In these studies, for *P. aeruginosa*, 13 of the 18 disinfectants passed at 20°C and 11 passed at 10°C, whilst for *E. coli* O157: H7, 15 and 14 disinfectants passed at the 20 and 10°C respectively. All disinfectants that passed for *P. aeruginosa* also passed for *E. coli* O157: H7.

It was concluded that *E. coli* O157 is not particularly resistant to disinfectants and that control of this organism within the factory environment should be possible with an effective disinfectant and sanitation programme.
Other authors have also studied the efficacy of disinfection regimes for *E. coli* O157. One of the first acidic products to be associated with contamination by *E. coli* O157 was apple cider and recent studies have looked at the effectiveness of a range of sanitising agents to inactivate *E. coli* on the surface of Golden Delicious apples (Sapers *et al.*, 1999).

It was found that washing apples with a 200ppm solution of chlorine, a typical disinfectant used for fruits and vegetables, achieved a 2-log reduction in levels of inoculated *E. coli* while the most successful treatment was the use of hydrogen peroxide at 50 °C which achieved a reduction of 3-4 logs. It is possible therefore that the use of effective decontamination procedures will help to eliminate the presence of any *E. coli* O157 in the food-processing environment.

**Conclusions**

Reported cases of food poisoning due to *E. coli* O157 are on the increase and the foods associated with this organism have diversified to include meats and meat products, fermented products, fruit and vegetables and mayonnaises. In order to control the contamination of foods or food environments with this organism, and the growth or survival during storage, it is necessary to have an understanding of the controlling factors for this organism in foods. Some of these controlling factors have been described in this paper and it has been shown that *E. coli* O157 is no more resistant than other organisms to temperature and disinfectants and therefore effective disinfection strategies and heat treatments should be effective against this organism. It has been shown that this organism can survive in acidic environments and the data described above have shown that careful choice of level and type of acid in combination with appropriate storage conditions can provide an effective control strategy.

**References**